REMARKS

Upon entry of this amendment, claims 1, 5-17, 19, 20, 24, 28, and 29 will be pending in the application. Applicants have canceled claims 2-4, 18, 21-23, 25-27, and 30 to expedite prosecution. Applicants reserve the right to prosecute claims of identical or similar scope in future applications.

The Examiner indicates that the Abstract was not received. Applicants hereby submit a copy of the Abstract page (which is page 94 of the specification as originally filed).

Applicants respectfully request reconsideration in view of the following remarks. Issues raised by the Examiner will be addressed below in the order they appear in the Office Action.

Claim rejections under 35 U.S.C. 112, first paragraph

Claims 1, 5-20, 24, 28 and 29 remain rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such as way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention. Specifically, the Office Action alleges that the specification does not contain an enabling disclosure for several reasons as elaborated below.

The Office Action asserts that the claimed invention is within the realm of an unpredictable and undeveloped art, and cited a passage from page 5, lines 1-10 of the instant specification to support this view:

> "...it has never previously been shown or suggested that treatment of myogenic precursor cells with the morphogens, morphogen inducers, agonists of morphogen receptors, or small molecule morphogenic activators is useful in promoting the proliferation and/or differentiation of myogenic precursor cells into new and functional myocardium in a morphogenically permissive environment. Nor has it previously been shown or suggested that morphogenically-treated myogenic precursor cells are useful in the treatment of lost or damaged mammalian myocardium."

However, Applicants submit that simply because something has never been shown or done before by another does not necessarily mean that the art is unpredictable. After all, any patentable invention has to be novel and unobvious in view of the prior art. It would not be an "invention" at all if someone has already shown or done something identical or similar before.

The reason why no one has done something similar or identical before is because the claimed invention is novel and unobvious, not because the art is so unpredictable that no one can do it. The difference between these two situations are best illustrated by the examples below. In the first scenario, someone may put a microphone on top of a pencil, so that he can simultaneously write out and dictate his ideas. If the pencil-microphone device is a novel and unobvious patentable invention, it may be because that no one has ever done or suggested this before, not because the art is so unpredictable that no one can construct a pencil-microphone. In other words, once the inventive idea is conceived, the rest is easy to implement based on the knowledge in the art. In the second scenario, before the invention of the Wright brothers, many attempts have been made by numerous others in trying to design a flying machine. However, in the highly unpredictable art of flying, it may well take the first successful flight of 100 or so yards, in addition to the blue prints of their aircraft on paper, to convince a patent examiner that their flying machine is a true invention but not a fantasy.

Applicants submit that the instant claimed invention resembles the first scenario described above. At the time of the claimed invention, the art was mature enough that, once a skilled artisan learned from the instant specification that morphogens can be used to treat myogenic precursor cells to promote proliferation or differentiation of these cells into functional myocardium, the skilled artisan would know how to isolate various types of precursor cells and treat them with morphogens prior to, simultaneously with, or after implanting them cells at the desired site. Furthermore, as argued in the last response, the instant specification also provides detailed information that will guide skilled artisans to practice the claimed invention.

Applicants' position is also well-supported by the case law. It is well-settled in case law that "[a]n inventor need not comprehend the scientific principles behind the invention. The inventor's theory or belief as to how his invention works is not a necessary element to satisfy the enablement requirement." *Cross v. Iizuka*, 753 F.2d 1040, 224 USPQ 739 (Fed. Cir. 1985). Therefore, Applicants need not describe any detailed mechanism behind the invention to enable the claimed invention. As long as Applicants have provided enough details (which Applicants have) as to how to make (how to administrate morphogens, etc.) and use (treat or prevent loss of myocardium) the claimed invention, the enablement requirement is met.

The Office Action also contends that "the specification does not teach the potential effects or morphogens on any kind of cells in any kind of biological activities," because

Applicants allegedly have not "provided art suggesting that any myogenic precursor cell or any morphogen can be used in the instant invention."

Applicants submit that in the experiment referred to in the specification (page 12, lines 10-14), at least one morphogen has been shown to promote the differentiation of precursors into functional myocardium. In addition, substantial post-filing scientific evidence, such as Behfar, proves that the claimed invention, if practiced as taught in the instant specification, would indeed work.

As to the useful morphogens, Applicants have amended claims 1, 15, 20, 24, and 28 to incorporate the subject matter of claim 18. As a consequence, claim 18 is canceled. Applicants submit that, according to Behfar, at least BMP-2 is highly effective in inducing precursor cell differentiation into functional myocardium. The BMP family of morphogens are highly related in structure and function to one another. The highly conserved C-terminal 6-7 Cys structure of these morphogens shares about 70% sequence homology to one another, while other more distantly related proteins, such as TGF-beta proteins, typically share less than 50% sequence homology. In addition, the BMPs / morphogens (but not other TGF-beta superfamily proteins such as TGF-beta, activin, or MIS) share the same set of cell surface receptors (type I and II BMP receptors) to exert their biological function. In fact, column 38 of U.S. Pat. No. 5,011,691 (issued on April 30, 1991) demonstrates that a few artificial sequences (COP-5 and COP-7) corresponding to the 6-7 Cys domain of the BMPs possess the same biological function (such as stimulating bone formation) as a full-length BMP protein OP-1/BMP-7. A skilled artisan, in view of this data, would readily understand that other morphogens within the scope of the amended claim 1 would also function similarly.

The Office Action raised several issues and suggests that **Exhibit A** (or "Behfar") filed with the last response is different from the subject matter sought to be patented. The concerns of the Office Action will be addressed below.

First of all, the Office Action asserts that Behfar cultures murine ES cell line in medium containing low serum and leukemia inhibitory factor (or "LIF"), and "differentiation was carried out in hanging drops of differentiation medium (20% FCS without LIF)." The Office Action thus contends that "the specification, however, as originally filed does not teach the use of LIF or any

other factor as suppressors of differentiation. The specification as originally filed does not teach specifically hanging drops or embryoid bodies as disclosed by Behfar."

Applicants submit that the experiments in Behfar sufficiently correspond to the teaching of the instant specification, and thus data obtained in Behfar constitute post-filing evidence that the claimed invention is enabled. On the other hand, the presence or absence of LIF, a standard medium component in this type of experiments, depends on the specific needs of the experiments and does not contradict any teaching of the instant specification. As a skilled artisan would appreciate, LIF was cloned in the late 1980's, and was known for a long time (before the earliest priority date of the instant application) as having a differentiation inhibitory activity in embryonic stem (ES) cells (see **Exhibit 1**, abstract of Williams *et al.*, *Nature* **336**: 684-7, December 15, 1988). In fact, the instant specification refers to LIF as having this inhibitory effect on page 17, line 3. As both Williams and Behfar indicate, if ES cells grow in a medium without LIF, the ES cells will randomly differentiate into a wide variety of cell types, see Figure 1d, second panel of Behfar. In contrast, ES cells grown in LIF-containing medium (see the "control" panel of Figure 1d) will not differentiate and will maintain the undifferentiated compact phenotype. Addition of BMP-2 to the LIF-containing medium would induce these ES cells to differentiate by initiating the "cardiac gene program" (Figure 1a-1c).

In an alternative experiment depicted in Figure 2, control cells are allowed to differentiate in medium without LIF, so that only a fraction of the differentiated cells become beating cardiomyocytes (see Figure 2b). In contrast, cells pre-treated with BMP-2, when put through the same differentiation process at the absence of LIF, form a three-fold larger contracting area compared to the untreated control (page 1561, left column, 1st paragraph).

Obviously, the presence or absence of LIF is being used according to the general knowledge in the art to control the outcome of the experiment. In this sense, LIF is no different from other factors required for cell growth in culture, such as FCS, pyruvate, supplimented non-essential amino acids, or mercaptoethanol (see page 1559, left column, second paragraph). No matter how LIF is used, the final conclusion of the Behfar experiments is that BMP-2 (and other related morphogens, plus TGF-beta) can stimulate ES cell differentiation into functional myocardium. Thus the specification need not teach the use of LIF, since it is immaterial to the invention sought to be patented, just like the other components of the medium used in Behfar are also immaterial to the claimed invention and need not be taught.

Similarly, the specification does not need to specifically teach the hanging drop differentiation of embryoid bodies as used in Behfar, since this is merely one of many possible models of verifying the conclusion described above. In fact, Behfar described another experiment in Figure 1, which also does not use this specific model. No matter which model is chosen, the conclusion is the same. The hanging drop differentiation method was at least described in a 1994 scientific publication (see reference 21 of Behfar – Maltsev *et al.*, *Circ. Res.* **75**: 233-244, August, 1994). It is just one way of allowing ES cells to differentiate *in vitro* under a controlled environment amenable to manipulation, and is just a routine way of running the experiment without critically affecting the final outcome.

A second concern of the Office Action is that Applicants have previously stated that TGF-beta is not a morphogen of the claimed invention, while Behfar demonstrates that TGF-beta has the same effect of BMP-2 in stimulating ES cells to become functional myocardium.

Applicants submit that this TGF-beta result does not conflict with the claimed invention. In fact, this result indicates that the specification has enabled beyond the claimed invention in terms of morphogen scope. The morphogens of the claimed invention are partially defined as sharing 70% sequence homology to the conserved C-terminal 6-7 Cys domain of human and mouse OP-1 and OP-2 (SEQ ID NOs. 4-7). As indicated above, TGF-beta only shares less than 50% sequence homology to the reference sequences, and thus does not fall within the scope of the recited morphogens. On the other hand, despite the low sequence homology between TGF-beta and the BMPs, TGF-beta still possesses the ability to promote myocardium formation, thus providing strong enabling evidence that other BMPs, which are substantially more closely related to BMP-2 than to TGF-beta, will also retain this biological function.

Thirdly, the Office Action is concerned that, unlike Behfar which uses an engineered cell line with a fluoresent marker protein (ECFP), the specification as originally filed does not teach the engineering of precursor cells to carry a marker demonstrating a specific phenotype. The Office Action is further concerned that the specification does not teach how to discern if the precursor cells have a cardiac phenotype.

Applicants submit that the reason Behfar uses ECFP as a marker in the engineered cells is to show that BMP-2 can induce the activity of cardiac-specific genes in the differentiating ES cells. In contrast, a patent application only need to teach a skilled artisan how to make and use

the claimed invention. In other words, while showing in the patent specification that the claimed invention would indeed work as said is desirable, it is not required. In fact, a patent specification can be based on prophetic examples, or contain no working examples at all, as long as a skilled artisan could make and use the claimed invention according to the specification. Applicants submit that the instant invention is partly based on the surprising discovery that morphogens can stimulate *in vivo* formation of functional myocardium (see page 12, lines 10-25). Thus, although not explicitly described, there is a working example which constitutes the basis of this application. However, even if there were no such working example, a skilled artisan could still make and use the claimed invention according to the instant specification, just as Behfar has done.

Relating to this, Applicants also submit that teaching a skilled artisan how to verify whether the precursor cells have become myocardium is not required to enable the claimed invention, since this is not one of the steps of the claimed invention, and is thus unnecessary for a skilled artisan to practice the claimed invention. The instant specification already states that morphogen treatment will stimulate myocardium formation, as subsequently verified by actual experiments conducted at least by Behfar. This demonstrates that a skilled artisan would be able to practice the claimed invention according to the specification without undue experimentation.

Lastly, the Office Action contends that the blocking experiments in Behfar (Figure 5) is not the same as implanting a myogenic precursor cell in a mammal, and treating the precursors with morphogen. "Behfar would need to show that injection of TGF-beta or BMP specifically into the implanted graft could rescue the phenotype i.e. express ECFP."

Applicants submit that the heart itself produces certain BMP proteins (see page 1561, right column, last paragraph), thus even without externally administered BMP proteins, the implanted precursor cells will be stimulated by these BMP proteins of endocrine origin to differentiate into functional cardiac muscle. It is to be expected that externally administered BMPs will enhance / accelerate this process. On the other hand, the experiment described in Figure 5 requires that no external morphogens be administered. This is because the experiment relies on seeing a difference in differentiation between cells that can respond to BMP, and cells that cannot respond to BMP due to the presence of inhibitors such as noggin (which binds and inactivates BMP, see Ref. 37 of Behfar – Zimmerman *et al.*, *Cell* **86**: 599-606). In the presence of a high concentration of externally administered BMP, the inhibitory effect of noggin would be

minimized, and there would be no expected difference in differentiation between these two types of cells. Thus the design of the experiment *mandates* the exclusion of externally administering morphogens in this particular experiment. Nevertheless, the result clearly indicates that it is morphogen, rather than other unidentified factors in the implanted heart, that stimulates the differentiation of the implanted precursors into functional myocardium.

The Office Action also asserts that page 39, lines 21-28, do not teach how one would treat precursor cells with a morphogen after the precursor cells have been implanted. "The specification fails to teach how to specifically target precursor cell once it is in the body."

Applicants submit that it is not necessary to specifically target precursor cells once they are in the body, just like most orally administered drugs do not act *exclusively* on their target cells / tissues. The morphogens can be administered systemically as taught on page 39, such as via *i.v.* injection. Since all blood passes through the heart (and thus the vicinity of the implanted precursor cells), morphogens administered that way would reach the implanted precursor cells. Alternatively, as taught on page 39, morphogens may be directly injected to the area where precursor cells are implanted. In fact, **Exhibit C** (Jackson *et al.*, *J. Clinical Investigation* **107**: 1395-1402, June, 2001) submitted with the last response indicates that stem cells do not even need to be implanted directly into the heart. The circulating hematopoietic stem cells can reach the heart and differentiate there to form functional myocardium. Although **Exhibit C** does not mention morphogen, it suggests that a similar technique can be employed to practice the claimed invention, by additionally administering morphogens (systemically or locally) to the mammal.

The Office Action also alleges that the specification fails to disclose and/or teach specific assays, materials, methods, etc. needed for the invention to be enabled. Applicants reiterate that assays to *verify* the differentiation of precursors are *not* required to enable the claimed invention since it is not part of the claimed invention. All other related knowledge, such as isolation of different types of stem cells, their implantation into damaged tissues, and administration of morphogens are all described in detail in the specification, and/or were well known in the art at the time of filing.

As to the use of skeletal muscle satellite cells, Applicants submit that these cells are multipotent and can commit to different differentiation fates (see also Wada *et al.*, Development 129(12): 2987-95, June 2002). There is no reason to doubt why this particular type of precursor

cell would behave differently than the tested stem cells in Behfar. Even if, for the sake of argument, the claimed invention were not enabled for satellite cells, there is ample evidence supporting the full enablement of adult stem cells or embryonic stem cells.

As to the timing of morphogen treatment, Figure 6 of Behfar indicates that precursor cells implanted *in vivo* into damaged (infarct) heart tissue will differentiate into functional myocardium in a BMP-signaling-dependent fashion (as proved by Figure 5), even in the absence of externally administered BMPs. This is presumably due to the presence of endocrine BMPs (see page 1561, right column, last paragraph). Thus it is conceivable that additional externally administered BMPs would further enhance this process. There is no compelling reason why external BMPs should need to be administered only before, during, or after the implantation of the precursor cells; thus, Applicants assert that all of these different embodiments are simultaneously enabled by the instant specification and verified by Behfar.

As to the morphogen fragments, Applicants reiterate that U.S. Pat. No. 5,011,691 indicates that two artificial polypeptides, COP-5 and COP-7, which correspond to the C-terminal 6-7 Cys fragment of the BMPs, and share just under 70% homology with the C-terminal region of the recited reference sequences, maintain the biological functions of the BMPs (see above). Thus the full scope of the claimed morphogens is enabled.

In summary, Applicants submit that the instant specification teaches in detail all necessary information for a skilled artisan to make and use the claimed invention, with no need to resort to any undue experimentation. Thus all pending claims fully comply with the enablement requirement of 35 U.S.C. 112, first paragraph. Reconsideration and withdrawal of the rejection are respectfully requested.

CONCLUSION

In view of the foregoing amendments and remarks, Applicants submit that the pending claims are in condition for allowance. Early and favorable reconsideration is respectfully solicited. The Examiner may address any questions raised by this submission to the undersigned at 617-951-7000. Should an extension of time be required, Applicants hereby petition for same and request that the extension fee and any other fee required for timely consideration of this submission be charged to **Deposit Account No. 18-1945.**

Respectfully Submitted,

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